

TRANSMISSION OF THE ROSE ROSETTE DISEASE AGENT TO *ROSA MULTIFLORA* BY *PHYLLOOPTES FRUCTIPHILUS* (ACARI: ERIOPHYIDAE)^{1,2}

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ABSTRACT: Tests proved that rose rosette disease (RRD) results from a pathogenic agent and is not a mite-induced response of multiflora rose. Graft transmission of the RRD agent to rooted cuttings of *Rosa multiflora* required 45 to 80 days and was only 46% successful. Graft transmission to large vigorously growing transplants was more rapid (30-60 days) and 100% successful. Graft transmission showed the agent resides in roots of multiflora rose. Laboratory transmission of RRD by the eriophyid mite, *Phyllooptes fructiphilus* (Acari: Eriophyidae) to transplants in 1986 was 92.3% and symptoms appeared in 17-24 days. Transmission of RRD by mites to rooted cuttings was unsuccessful. In 1987, the rate of field transmission with *P. fructiphilus* was 12.5% and lab transmission was 20% with symptoms appearing in 30-279 days and 29-47 days respectively. Reduced laboratory transmission in 1987 was thought to be drought-induced. Attempts to transmit RRD with *Tetranychus urticae* (Acari: Tetranychidae) were unsuccessful.

Rose rosette disease (RRD) affects numerous rose species, especially *Rosa multiflora* Thunb., and has been reported in the midwestern states (Allington *et al.* 1968, Crowe 1982 and 1983, Doudrick and Millikan 1983, Gergerich and Kim 1983, Gergerich *et al.* 1983, Hindal and Amrine 1987 and 1989). The nature of the causative agent remains unknown but studies suggest it may be a virus (Gergerich and Kim 1983) or a mycoplasma-like organism (Doudrick 1984).

The causal agent of RRD has been graft-transmitted (Thomas and Scott 1953, Allington *et al.* 1968, Doudrick 1984). Allington *et al.* (1968) demonstrated transmission by the eriophyid mite, *Phyllooptes fructiphilus* Keifer (Fig. 1), to *Rosa eglantheria* L. (16.7%) and to *R. multiflora* Thunb. (34.3%) using mites collected from several species of roses showing symptoms of RRD. Allington *et al.* (1968) demonstrated that RRD was not a mite-induced plant reaction while Slykhuis (1980) suggested

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that RRD may be the result of a toxicogenic reaction to mite feeding. Doudrick *et al.* (1986) failed to obtain transmission by the mite when placed on rooted cuttings of a healthy thornless clone of *R. multiflora*.

We present results of experiments of RRD transmission by grafting and mite feeding that prove 1) RRD is an infective agent of multiflora rose and 2) that *P. fructiphilus* can transmit the RRD agent to multiflora rose. We also demonstrate retention of the agent by *P. fructiphilus* and discuss variables that may influence transmission trials.

MATERIALS AND METHODS

Plant material used in transmission tests

Rooted cuttings and pruned, transplanted field grown healthy multiflora roses (transplants) were used in these tests. Stems from the thorny variety of multiflora rose were cut into three-node sections, their bases coated with Rootone (R) and placed in a greenhouse mist bed for 4-6 weeks. When rooted, these cuttings were placed in a peat-vermiculite (1:1) mix in four inch pots. The transplants were obtained by pruning healthy field grown multiflora rose plants (from the vicinity of Morgantown, West Virginia) until the stems were about 30 cm long; the plants were then dug and transplanted into 30 cm pots containing the above

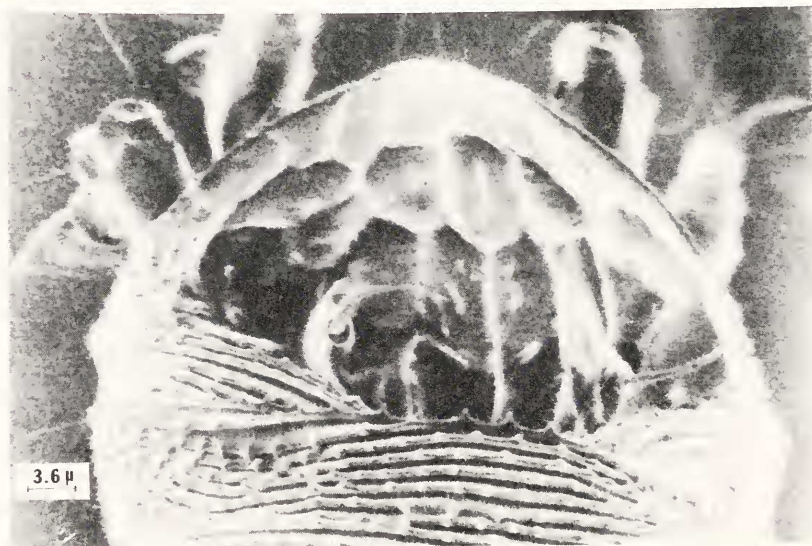


Figure 1. SEM micrograph of dorsal shield of *Phyllocoptes fructiphilus* K.

mix. All cuttings and transplants were maintained in a greenhouse containment room. Plants were sprayed with dicofol (Kelthane), propargite (Omite), oxydemetonmethyl (Metasystox R), and cyhexatin (Plictran) as needed to control spider mites (*Tetranychus urticae* Koch) and fertilized monthly with Peter's Professional, General Purpose 20-20-20 fertilizer at 1.3 ml/l monthly.

Graft and mite transmission

Rose rosette disease infected tissues were obtained from multiflora rose shoots showing symptoms of RRD collected in S.E. Missouri, W. Kentucky and southern Indiana (Hindal and Amrine, 1987, 1989). Four graft transmission experiments were conducted with these tissues. Rooted cuttings were used in experiments 1 and 2 and transplants with succulent regrowth were used in experiments 3 and 4 (Table 1). In experiments 1, 2, and 3, pieces of infected stem tissue (3 to 5 mm long), and free of eriophyid mites were bud grafted (one per stem) onto at least three stems of each test plant. In experiment 4, eight pieces of tissue from a 5 mm diameter root of an infected plant showing symptoms of RRD were bud grafted into each of four plants. Tissues from Missouri and Kentucky were used in experiments one through three, whereas tissues from Southern Indiana were used in experiment 4.

In transmission tests using the mite, *P. fructiphilus* (Fig. 1), cuttings, transplants and field plants were used as bioassay plants. For mite transmission experiments 5 and 6, mites were field collected from *R. multiflora* showing symptoms of RRD from Scott County, Indiana on 12 Aug. 1986. These were stored at 1°C for nine days. For mite transmission experiments 7 and 8, mites were collected on 26 Sept. 1986 from affected plants at Madison Indiana and stored at 4°C for three days (Table 2). For field experiments 10 through 21, mites were collected on dates indicated in Table 3 at Clifty Falls State Park the day before experiments were conducted. For laboratory experiments 22 through 25 in 1987 (Table 4) mites were obtained from Clifty Falls State Park and for experiment 26, from Caesar Creek State Park, Warren County, Ohio.

In all mite transmission experiments, leaves of shoots showing symptoms of RRD were trimmed just above stipules and the petioles examined microscopically for mite populations. Only those petioles with 10 or more living mites were used. Infested petioles containing ca. 10 mm of stem were cut from the stems and placed in the apical leaf axils of the test plants. This procedure placed the mites into direct contact with their preferred developmental site (i.e., the petiole and axillary bud) on the test plant. The natural tendency of each leaf petiole to press against its stem acted as a double clamp, holding the petioles together.

Only one trial, experiment 5, was conducted with rooted cuttings. These cuttings were 3 months old and contained two or three small branches, but were not rapidly growing. The other laboratory mite transmission tests were conducted with transplants containing 15 to 30 cm of regrowth. Experiment 9 was conducted with "progeny mites" that had developed on the plants from experiment 8. These progeny mites on petioles were removed ten days after initiation of experiment 8 and placed, three petioles each, onto ten transplants (Table 2). In all mite transmission experiments, growing shoots were removed and examined for the presence of *P. fructiphilus* seven days after the mite-infested petioles were placed on bioassay plants for laboratory experiments, and after 30 days in field experiments. The presence of mites (especially of eggs and immatures) indicated that mites were successfully established on test plants. All plants in the laboratory transmission tests were treated with dicofol (Kelthane) and aldicarb (Temik) no later than 14 days after inoculation to eliminate both eriophyids and spider mites. In two of the field transmission experiments, one mite (experiment 11) and four mites (experiment 14) were placed onto each of 20 small leaf pieces which were then placed into growing tips of test plants to observe whether individual mites or a few mites could transmit RRD. In two of the 1987 laboratory experiments (22 and 24), *P. fructiphilus* were maintained on infected tissue for two weeks at 4°C to check for retention of the agent. (At this temperature, *P. fructiphilus* is inactive and feeding does not occur (Amrine, unpublished)).

After the transmission trials were initiated, the time, appearance, and number of plants showing RRD symptoms were observed. Test plants were determined to be infected when characteristic red or purplish pigment appeared in spots or blotches on the leaves (spot mosaic) or, later, when the veins became strongly pigmented with red (vein mosaic) (Fig. 2). These symptoms in combination with the later appearance of bright red shoots developing from the dormant buds at leaf axils, indicated that the RRD agent had been successfully transmitted (Amrine and Hindal, 1988).

During 1985 and 1986, 36 healthy multiflora rose plants in the field near Morgantown, WV, were artificially infested with *P. fructiphilus* collected from non-symptomatic plants in Cabell County, WV. These eriophyids were morphologically identical to mites collected on multiflora rose plants with RRD symptoms in Missouri, Kentucky, Indiana and Ohio.

Ten transplants were placed in a cage around a diseased plant infested with *T. urticae* (experiment 27). Canes from the symptomatic plant were placed so that they were in contact with each test plant to insure passage of spider mites. The plants were kept together for 30 days then treated with dicofol and aldicarb to eliminate the mites; during this time,

very large numbers of *T. urticae* developed and migrated among all of the plants.

RESULTS

Graft transmission in rooted cuttings. Grafting experiments in rooted cuttings showed slow development of RRD and low transmission rates. In experiment 1, a total of five of 12 plants (41.7%) showed symptoms of RRD within six months. In experiment 2, symptoms appeared as follows: four of 37 (10.8%) in 41 days, six of 37 (16.2%) by 51 days, and 17 of 37 (46%) by 80 days. Table 1 summarizes the results of the experiments.

Graft transmission in large transplants. In experiment 3, 100% of the large plants showed symptoms of RRD within 60 days of grafting. In experiment 4, all four plants grafted with infected root tissue showed symptoms in 75 days.

Mite Transmission. In experiment 5, no mites were recovered from the rooted cuttings and no symptoms developed. In experiment 6, the



Figure 2. The two leaves at the left show "spot mosaic", the first symptoms of rose rosette disease; the leaf at right shows typical vein mosaic (deep red to maroon), a characteristic advanced symptom of rose rosette disease.

plants were examined after seven days and developing mites (eggs + nymphs) were found on three of eight plants, indicating the successful establishment of mites on 37.5% of the trial plants. Four plants (50%) showed symptoms of RRD in 24 days. A fifth plant (62.5%) developed symptoms in 30 days while a sixth plant developed typical symptoms after 160 days. This indicated an overall infection rate of 75%. In experiment 7, four of eight plants contained developing mites after 10 days, thus mite establishment was successful on 50% of the plants. Six plants (75%) showed symptoms in 17 days, and all 8 (100% transmission) showed symptoms in 30 days. In experiment 8, all shoot tips examined after 10 days contained developing mites (100% successful transfer) and all 10 plants developed symptoms in 17 days (100% transmission). In experiment 9, all plants had successful mite establishment and only one plant developed symptoms in 17 days (10% transmission). No additional plants became infected after 160 days. After elimination of mites with pesticides, RRD symptoms continued to develop, producing mosaic, red lateral shoots, and witches' brooms, and eventual death of plants.

Field transmission, 1987. In experiments 10-18, 13 of 104 plants became infected in 30-279 days for 12.5% transmission. Seven of the infected plants did not show symptoms until 26 April 1988. One plant in the single mite transmission experiment and three plants in the four mite transmission experiment showed symptoms on 26 April 1988, 279 days after initiation of the test. Neither of the transmission experiments (19 and 20) conducted in the field on 23 Sep. 1987 were successful, probably because temperatures were too low for mites to be active and to feed.

Laboratory transmission, 1987. In experiments 22-26, ten of 50 plants showed symptoms of RRD in 29-70 days for 20% transmission; for experiments 22 and 24, mites had been held at 4°C for 14 days, indicating retention of the pathogen by the mites.

In establishment of *P. fructiphilus* on healthy multiflora rose, none of the 36 large field plants showed symptoms of RRD or any symptoms of mite infestation. None of the plants fed on by *T. urticae* became symptomatic.

DISCUSSION

The slow and incomplete development of RRD symptoms in RRD-grafted rooted cuttings (a total of 22 of 49 plants or 44.9% developed symptoms in 70 to 80 days) was quite striking as compared to the RRD-grafted large transplants (100% in 60 days). The physiological or pathological basis for this difference is unknown. In contrast to the production of

numerous new shoots in the transplants, the rooted cuttings were not rapidly growing, and this difference may have affected the results: the causative agent of RRD may require rapidly growing tissue for efficient establishment. The high graft-transmission rate in transplants may have been enhanced by the stress condition of the plants (canes and roots were pruned and the plants were using stored reserves to produce new growth), rather than to differences in size or growth condition. It appears that transmission is incomplete in rooted cuttings and large transplants are better hosts for the identification of RRD.

The development of RRD symptoms from grafting of mite-free, symptomatic tissue supports our hypothesis that RRD is caused by a pathogenic agent and does not reflect a reaction to feeding of the eriophyid mites. The successful transmission of RRD by grafting of root tissue from RRD affected plants to stems of healthy plants (experiment four) also supports this hypothesis, since no eriophyid mite has been recovered from root tissue (Keifer, 1975). This last graft trial also indicates that the agent for RRD resides in the roots of *R. multiflora*.

During the past two years we have kept more than 200 transplants in a separate room as reserves for experiments and for future maintenance of the RRD agent. During this time, none of the plants developed symptoms of RRD, indicating that RRD was not present in field plants in West Virginia. Thus, positive results in our experiments could not have resulted from contamination or inapparent infection of dug field plants.

Definitive conclusions about the transmission of a disease agent by eriophyid mites requires that the mites must be successfully established on the test plants. *Phyllocoptes fructiphilus* requires succulent, growing tissues near apexes of shoots for successful feeding and development (Amrine and Kharboutli, unpublished). In experiment 5, the rooted cuttings were not growing rapidly and apparently were unsatisfactory for mite establishment. Failure to find any mites on the plants after seven days and the failure of RRD symptoms to develop suggests that the mites were unable to feed on the cuttings.

In experiments 6 through 8, establishment of *P. fructiphilus* on the new shoots was 92.3% successful, and RRD symptoms appeared on 18 plants by 17 days. These trials displayed the full potential of *P. fructiphilus* as a vector of the RRD agent to multiflora rose.

In experiment 9, only one plant showed symptoms of RRD in 17 days and no additional plants developed symptoms after more than 160 days. Apparently, *P. fructiphilus* loses much of its capacity to transmit the RRD agent after 10 days. Since it could not be determined whether "progeny" mites were indeed all progeny or were populated by some of the original mites, we can not determine the mechanism of this 10% transmission. The RRD agent may have just begun to multiply in stems and leaves of

the newly infected plants, or a few original mites may have still been present on the transferred petioles.

The appearance of RRD symptoms within 17 days in experiments 7, 8 and 9 was the most rapid appearance of symptoms of RRD reported to date in the literature. Allington *et al.* (1968) reported the development of symptoms in 30-103 days. It appears in our investigations that *P. fructiphilus* (under optimum conditions) is more effective and efficient in introducing the RRD pathogen to susceptible tissue compared to graft transmission. The selection of healthy, large plants, dug and pruned two to four weeks before the experiments and growing vigorously at the time of the trials, was critical to the success of our experiments, both for grafting and mite transmission. The preference of *P. fructiphilus* for feeding on rapidly growing tissue near shoot apices (where cells are very small and the 10-20 micron stylets may be able to extend past the epidermis) may give the RRD agent direct access to the xylem, phloem, or other specific host tissue which may harbor or support the RRD agent. The slower developing grafts may require longer periods of time to develop contact with these tissues. Size, growth condition, vector site preference and stress of host plants may affect the results of transmission tests of other suspected, or known, eriophyid transmitted disease agents and should be considered when conducting such tests.

Success of field and laboratory transmission trials with *P. fructiphilus* in 1987 were much less than the 92.3% successful laboratory trials conducted in 1986. Southern Indiana endured a moderate to severe drought in 1987; precipitation from April through September (14.4") was only 62% of normal (23.2") (Scheeringa, 1987). The drought affected both healthy and diseased plants of *R. multiflora* and resulted in greatly reduced new growth, and much of the foliage turned yellow and dropped off. The drought may have affected the transmission trials by modifying the mites' development or their ability to transmit the pathogen, or by reducing the availability of the pathogen in plant tissues to the mites. The drought also may have delayed expression of RRD symptoms in field plants which was evident in the appearance of some infections in April 1988. These results indicate the variability of mite transmission from season to season and the need for continuing thorough studies of all aspects of eriophyid transmission of RRD.

Transmission experiments employing one mite (Experiment 11) and four mites (Experiment 14) were successful, but symptoms did not appear until 26 April 1988. These two experiments indicate that adult *P. fructiphilus* can transmit RRD, and that single or a few adults can transmit the disease. Likewise, it proves that the mites are able to leave small pieces of drying leaves and cross onto healthy plant tissue. These two experiments also indicate that the field plants were slow in develop-

ing symptoms, perhaps because of the drought, or perhaps because a very small amount of inoculum was transferred (i.e., the larger the number of infected mites feeding, the more inoculum delivered, and the more rapid the development of symptoms). More likely, the drought retarded appearance of symptoms, since plants in four petiole experiments (one each in experiments 10, 13, 16 and 18) also did not develop symptoms until 26 April 1988.

In all of the mite transmission experiments, recovery of mites from test plants after seven days was variable. In many cases mites were found on tips but symptoms did not develop, and in other cases (experiments 6, 7, 14), symptoms developed but mites were not recovered. We believe that these discrepancies may result from two possibilities. For one, only a fraction of the mites may carry the RRD agent or be able to transmit it. And two, the mites may feed and transmit the agent, but for some reason not always establish a colony. The failure of the 36 large *P. fructiphilus* infested multiflora plants near Morgantown to develop any symptoms also indicates that RRD is not a mite-induced host response. Furthermore, during 1985 to 1987, several hundred multiflora rose plants were found infested with *P. fructiphilus* in eastern KY, OH, MD, NC, SC, and WV, but none showed symptoms of RRD. As of May 1988, no multiflora rose in West Virginia have been found to show symptoms of RRD.

Tetranychus urticae is not able to transmit RRD (Experiment 27). This pest has been a severe problem in our greenhouse; however, no unexplained transmission of RRD has appeared that may have resulted from transmission by *T. urticae*.

The relationship of the pathogen to the vector is of major importance to transmission of RRD by *P. fructiphilus*. Is the RRD agent merely a contaminant on the mouthparts or in the digestive tract, or does it actually penetrate the gut epidermis and eventually infect salivary glands? Infection of salivary glands has been demonstrated for *Aceria tulipae* K., which transmits Wheat Streak Mosaic Virus to wheat (Paliwal, 1980). Retention of the RRD agent in *P. fructiphilus* held on infected tissue for 14 days at 4° (experiments 22 and 24) suggests that the agent may be semipersistent or persistent, but more work needs to be done to elucidate this problem. It is unknown why mites held at 4°C for 7 days failed to transmit RRD (experiment 23) while retention for 14 days succeeded. We believe that because of the low rate of transmission (20%) that it is not statistically unusual to obtain no results in one test of 10 plants.

This study corroborates the conclusions by Allington *et al* (1968) that RRD is an infectious agent of multiflora (and other) roses and that it is transmitted by the eriophyid mite, *P. fructiphilus*. We believe that Doudrick *et al*. (1986) were unable to prove transmission of RRD by *P. fructi-*

iphilus because of the succulent tissue feeding requirement of the mites and also the incomplete transmission of RRD to rooted cuttings versus larger transplants. This study also shows that transmission of RRD by *P. fructiphilus* can be erratic and affected by drought or plant stress or both.

Mites collected on 20 October and 9 November were able to transmit RRD in 39 and 33 days respectively in the laboratory (experiments 25 and 26). However, petiole trials conducted in the field on 23 September (experiments 19-21) failed to show transmission. Experiment 20 employed 3 petioles with mites per plant, yet proved negative in transmission. We therefore believe that either the mites were unable to feed in the field after 23 September due to low temperatures, thus transmission could not occur, or that plants in the field could not be infected after August, perhaps due to changes in physiology as autumn and winter approaches.

Related to this discussion were collections of *P. fructiphilus* on *R. multiflora* in Fayette Co., WV, on 6 December 1985, when very large populations of mites were found, including large numbers of eggs and immatures. Also, many of the plants had developed succulent new growth at this late date because of the unusually warm fall weather. In contrast, most of the multiflora rose in Indiana and WV in fall 1987 had lost foliage and become hardened off by October. The considerable variation in growth condition of *R. multiflora* from one year to another is also reflected in potential transmission. We believe that when field moisture is adequate, temperatures are warm, and *R. multiflora* produces copious new growth, that mite populations and hence the potential for transmission of RRD are correspondingly high.

Studies in our laboratories will continue to examine the nature of *P. fructiphilus* transmission of RRD. Emphasis will be made on EM examination of *P. fructiphilus* tissues from both RRD affected and healthy plants, comparison of immature and adult mites in transmitting RRD, "feeding" times required to achieve acquisition and inoculation of the agent, retention of the agent by mites, and effect of stress on host plants to mite biology and mite transmission of RRD.

Table 1. Graft transmission of Rose Rosette Disease to *Rosa multiflora* rooted cuttings and transplants.

Experiment No.	Inoculum ¹	Bioassay type	Grafts		Development of Symptoms		
			No. of Plants	Date	Shortest Incubation Period (days)	No. Plants with Symptoms	% Infection
1	stem	cutting ²	12	24 Dec 85	80	5	41.7
2	stem	cutting ³	37	30 Jul 86	41	17	46.0
3	stem	transplants ⁴	30	Dec 85	30	30	100.0
4	root	transplants ⁴	4	20 Feb 86	53	4	100.0

¹From plants showing symptoms of RRD.

²From rooted 3 node sections of healthy multiflora rose, growth rate slow.

³From rooted 3 node sections of healthy multiflora rose, growth rate moderate.

⁴From large, pruned field plants, dug and transplanted, growth rate rapid.

Table 2. Laboratory Transmission of Rose Rosette Disease by *P. fructiphilus* to cuttings and transplants of *Rosa multiflora*, 1986.

Experiment No.	Bioassay type ²	Mite Transfer		Shortest Incubation Period (days)	No. Plants with Symptoms	Plants with Mites
		No. of Plants	Date			
5	cuttings ³	10	21 Aug 86	—	0	0
6	transplants ³	8	21 Aug 86	24	6	3
7	transplants	8	29 Sept 86	17	8	4
8	transplants	10	29 Sept 86	17	10	10
9	transplants ⁴	10	9 Oct 86	17	1	10

¹Mites were transferred from an infected multiflora rose showing symptoms of RRD. Each petiole contained 10 or more mites. Infected petioles for Experiments 5 and 6 originated near Scottsville, IN. Infected petioles for experiments 7 and 8 were obtained from Clifty Falls State Park, Madison, IN. Mites (10 or more per petiole) for experiment 9 were collected from plants in Experiment 8, ten days after original transfer.

²Cuttings were 3 months old, with two or three branches and growth was slow. Transplants were healthy, large, pruned field plants, dug and transplanted 14 to 30 days before the experiment, and growth was rapid; with new stems 10 to 30 cm long.

³Treated with dicofol on 11 Aug. 1986 to control spider mites.

⁴Treated with dicofol on 7 Sept. 1986 to control spider mites.

Table 3. Field Transmission of Rose Rosette Disease by *P. fractiphilus* to Multiflora Rose in Southern Indiana, 1987.

Experiment	Inoculum Type	Location	Date	No. of Plants	Development of Symptoms ²											
					Transfer of Mites			16 Aug 87			22 Sep 87			19 Oct 87		
					S	M	S	M	S	M	S	M	S	M	S	
10.	petioles ¹	Snyder Prop.	21 Jul 87	20	1	15	3	-	4	-	-	-	-	-	5	
11.	1-mite	Snyder Prop.	21 Jul 87	20	0	1	3	-	4	-	-	-	-	-	1	
12.	control	Snyder Prop.	21 Jul 87	20	0	1	0	-	0	-	-	-	-	-	0	
13.	petioles	Rexville	23 Jul 87	20	0	2	1	-	1	-	-	-	-	-	2	
14.	4-mites	Rexville	23 Jul 87	19	0	2	0	-	0	-	-	-	-	-	3	
15.	control	Rexville	23 Jul 87	20	0	0	0	-	0	-	-	-	-	-	0	
16.	petioles	Versailles	18 Aug 87	20	-	-	0	14	0	-	-	-	-	-	1	
17.	control	Versailles	18 Aug 87	10	-	-	0	0	0	-	-	-	-	-	0	
18.	petioles	N. of Rt. 50	18 Aug 87	5 ³	-	-	1	3	1	-	-	-	-	-	1	
19.	petioles	Versailles	23 Sep 87	20 ³	-	-	-	-	0	-	-	-	6	-	0	
20.	3-petioles	Snyder Prop.	23 Sep 87	20	-	-	-	-	0	-	-	-	9	-	0	
21.	control	Snyder Prop.	23 Sep 87	10	-	-	-	-	0	-	-	-	0	-	0	

¹Mites from infected plants (Clifty Falls, St. PK.) were placed into growing tips of test field multiflora rose as follows: "petioles" were removed from infected plants and contained 10 or more mites; "one-mite" was single, adult mites from an infected rose placed onto a small piece of leaf; "four-mite" was four adult mites from an infected rose placed onto a small piece of leaf.

²On dates listed, plants were examined for number developing symptoms (S) and number with mites present (M).

³Test plants were cut in August to test for effect of stress on transmission.

Table 4. Laboratory Transmission of Rose Rosette Disease by *P. fructiphilus* to transplants of *R. multiflora*, 1987.

Experiment	Mites			Development of Symptoms				
	Date of Collection ¹	Exposure to 4°C (days)	Source	No. of Plants	Date of Inoculation ²	Length of Incubation (days)	No. Plants with Symptoms	No. Plants with Mites ³
22	19 Aug	14	Clifty Falls, IN	10	2 Sept	47	2	10
23	22 Sept	7	Clifty Falls, IN	10	29 Sept	—	0	7
24	22 Sept	10	Clifty Falls, IN	10	6 Oct	29	2	9
25	20 Oct	3	Clifty Falls, IN	10	23 Oct	39	3	5
26	9 Nov	1	Caesar Creek, OH	10	10 Nov	23	3	6

¹Mites on symptomatic shoots were kept in plastic bags in a dark refrigerator at 4°C until transmission tests were conducted.

²Three petioles, each containing at least 10 mites, were transferred to indicator plants.

³Three tips examined on each indicator plant.

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